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15. ABSTRACT Most chlorine toxicity studies have been conducted under conditions of constant exposure, rather than under intermittent chlorination conditions. The recent emphasis to generate more appropriate data under power plant operational modes will result in a more comprehensive and appropriate basis for criteria development. In the interim, several considerations need to be discussed in detail: (1) toxicity is time, concentration, water quality, and species related; (2) disequilibrium usually results in mortality; (3) various physical, biological, and chemical factors may influence observed effects at plants during chlorination; and (4) a representative research program involves variable exposure times, exposure concentrations, water quality, both fish and invertebrate species.					
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General Considerations Concerning the Toxicity to Aquatic Life of Chlorinated Condenser Effluent

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INTRODUCTION

Most of the early studies on the toxicity of chlorine were conducted under static bioassay conditions, the standard procedure in use at that time. The results of those studies gave the false conclusion that chlorine was not very toxic to aquatic life. In reality, however, the high concentrations required to kill the test organisms were the result of fairly rapid detoxification which would occur under the static conditions. Later, when continuous-flow testing became common, most investigators adopted those procedures. As a result the toxicity literature contains the results of many studies that indicate the very toxic conditions caused by chlorination of waste effluents. Some of these studies were conducted using chlorinated wastewater effluent from sewage treatment plants. Others used the classical toxicity test using clean water under laboratory conditions. Both usually involved continuous (and constant) exposure for the usual test duration of 96 hours; some chronic studies demonstrated very low "safe" concentrations. It has been only recently that anyone was concerned about the chlorination problem as it related to the power generation industry. Much of this concern was probably the result of limitations being placed on the industry as part of the USEPA effluent permit program and various recommended aquatic life criteria for intermittent chlorination (Basch and Truchan 1974; Brungs 1973). At the present time there are several studies under design or being conducted that are specifically oriented to the needs of the power generating industry and there should be sufficient data within one to two years to evaluate and/or expand the present criteria.

FACTORS THAT INFLUENCE TOXICITY

Probably the most important aspect of water quality effects on toxicity is its influence on the ratio of free chlorine and combined chlorine (chloramines). This ratio is important, since available data indicate that free chlorine

is more toxic and/or more rapidly toxic than combined chlorine. Truchan has described field studies at various Michigan power generating stations at which free chlorine was detected for at least several hundred yards downstream. These trout waters had a low chlorine demand. While such a short distance may seem unimportant in relation to size of the thermal plume, we must remember that it is in this relatively small area that large numbers of fish may congregate during cooler seasons. At most sites the concentration of free chlorine will be insignificant, but if chlorine is being applied at an unnecessarily high concentration the likelihood of free chlorine in the discharge will increase because the chlorine demand of the cooling water is being exceeded.

Several recent studies have attempted to determine the influence of temperature on the toxicity of total residual chlorine. Truchan has done studies that have shown a slight increase in toxicity with an increase in temperature and Seeger has observed a difference of about 7% in LC50 values for yellow perch over a temperature range of 10-20 deg. C. He also indicated, however, that the LC50 values for coho salmon at 10 and 15 C were 1.25 and 1.3 mg/l, respectively; this would indicate little if any effect. An excellent study by Gregg (1974) with stream invertebrates, such as mayflies, stoneflies, snails, cruds, and other larval insects, has shown that for some species temperature caused a small increase in the toxicity of intermittent exposure to chlorine and for other species there was no change in sensitivity. The influence of temperature on the toxicity of various chemicals, including chlorine, was discussed in a recent review by Cairns et al. (1975). He thought that if chlorine affected oxygen transfer across the gills an increase in respiratory demand for oxygen due to increased temperature could increase the toxicity of chlorine to fish. I would conclude that in most instances the influence of temperature on chlorine toxicity is slight, and additional thorough experimentation is necessary.

The present published literature contains many data that force the conclusion that trout and salmon are more sensitive to total residual chlorine than are the warmwater fish

species. That was the basis for a 5M difference in the recommended criteria for these species groups for continuous and intermittent chlorine discharge. Much of the information on invertebrate species indicated that many were very resistant to chlorine and only a few were as sensitive as the trout and salmon. A study at the Grandville Wastewater Treatment plant in Michigan, sponsored by the USEPA, will provide much additional information on many additional fish species. This study has indicated that for continuous 96-hour exposures to chlorinated wastewater effluent, some species of minnows important as forage fish appear to be as sensitive as the salmonids. Data on a variety of invertebrate species (Glegg 1974; Grossnickle 1974) indicate that some aquatic insects and rotifers are also as sensitive as the salmonids. Many aquatic invertebrates, however, appear to be quite resistant to total residual chlorine (Glegg 1974; Latimer 1975). These latter studies included data on intermittent exposure to chlorine. If these data for minnows and some fish-food organisms are supported by additional studies, it may become necessary to eliminate the distinction between warmwater and coldwater fish.

For most, if not all, toxicants aquatic life responses are time/concentration related. This means that the longer an animal is exposed to a stress, the lower that stress level must become to permit survival. Conversely, aquatic life could, within limits, survive higher concentrations of a stress if the length of exposure were shortened. As additional field and laboratory data become available it is possible that chlorine criteria may become time-dependent, with several concentrations of chlorine for different chlorination times. We must remember, however, that a 30-minute chlorination results in an increasingly longer exposure time as the slug passes downstream from the discharge point.

DIS-EQUILIBRIUM AND AVOIDANCE

Mortality as a visual endpoint of a toxic response is not adequate to describe the real impact of intermittent exposure to chlorine. Dis-equilibrium, or loss of balance, may be the point of no return for a chlorine-disabled fish. Dandy (1973) determined that when brook trout were transferred directly into freshwater at the first signs of dis-equilibrium caused by chlorine, death always ensued, even though life expectancy at the time of transfer from lethal solutions was several hours. Pike (1971) observed essentially the same responses after transfer of affected brown trout to clean water. Studies that only determine mortality as the endpoint for fish after exposure to intermittent chlorine may underestimate the real impact of eventual mortality after the exposure terminates. In addition to mortality, dis-equilibrium should also be used as an endpoint in chlorine toxicity tests. The term *effective concentration* (EC) has been proposed in connection with

effects other than death (American Public Health Association 1971). Therefore, a concentration of residual chlorine that results in 50% of the test fish losing equilibrium would be the EC50. Once fish have lost their equilibrium they are subject to greater predation pressure, and gulls have been observed actively feeding on smaller fish forced to the surface during a chlorination period (Beach and Truchan 1975).

Some believe that the impact of power plant chlorination has been exaggerated by assuming that most of the fish will not actively avoid lethal concentrations. I am aware of only a few studies that attempted to evaluate this condition. River shiners, bullhead minnows, and mimic shiners were shown to avoid gradually increasing concentrations of total residual chlorine in the range of 0.005 to 0.45 mg/l with the mimic shiner being the most sensitive (Bogardus et al. 1975). Sprague and Drury (1969) observed that rainbow trout significantly avoided 0.01 mg/l of chlorine which would be lethal in 12 days and 1.0 mg/l which was rapidly lethal. Most of the fish preferred an intermediate lethal concentration of 0.1 mg/l. Both studies were conducted in avoidance chambers that provided a distinctly and readily available control area and the fish were always within a fraction of a meter of that safe area. In a study (Meldrim et al. 1974) with marine species, the mummichog, hogchoker, grass shrimp, and sand shrimp did not demonstrate a preference for a lethal concentration of chlorine as did the rainbow trout. However, the blue crab did demonstrate a preference for lethal conditions. The authors concluded that, although the data were too sparse for statistical analysis, avoidance concentrations were inversely related to temperature and to light level. The effect of reduced oxygen on chlorine avoidance varied slightly by species. Reducing the level of dissolved oxygen increased the avoidance concentration of the hogchoker, but usually reduced it for the other species.

At power plant discharge areas, distance and time need to be considered also. If the fish which would attempt to avoid the chlorine slug by random directional movements could not find a safe area within a short time period before they lose equilibrium, the avoidance phenomenon would have no positive survival value.

COMBINED EFFECTS

The synergistic or additive effect of combined stresses should be considered when evaluating the potential impact of chlorination. Chlorine criteria cannot and do not include any safety factor for these other stresses (McLean 1973; Stober and Hanson 1974; Dressel 1971; Hoss et al. 1974; Gibson et al. 1975; Thatcher et al. 1975; Wolf et al. 1975). The most important fact to remember is that if chlorine criteria are being consistently met or slightly exceeded in power plant effluents, the additional stresses that periodically occur may result in an unexpected fish kill.

Temperature increase may affect the incidence of infectious diseases of fish. Saito (1974) reviewed the effects of increased temperature on the occurrence of outbreaks of bacterial and viral diseases and concluded that water temperature may affect these diseases if the potential pathogens are present. Mortality rates for salmonids due to *Chondrocytes columnares*, *Aeromonas salmonicida*, and *Aeromonas hydrophila* were increased with an increase in temperature over the range of 4-21 °C (Fryer and Pitcher 1974). Mortality of coho salmon exposed to *Ceratomyxa shasta* was also increased with increasing temperature. A disease stressed fish is probably more sensitive to chlorine. This condition would not occur at all times but the possibility does exist and is one that is unlikely to be studied in the laboratory.

Vaughan and Cable (1975) have evaluated the influence of ectoparasites on the resistance of fish to thermal stress. They found that brook trout with gill lice (*Salmicola edwardsii*) had a 50% mortality within 32 to 33 hours at an exposure of 35 °C. Trout without gill lice did not reach a 50% mortality during the experiment. They also found that yellow perch with black-spot (*Neascus of Crassiphiala hubbsii*) were actually more resistant in relation to survival time at 33 °C.

Some aquatic environments still have relatively high quality water in association with power generating stations. In these situations, where the usual dissolved gas concentrations are at or near saturation, heating the water will result in supersaturation of these dissolved gases. At the Waukegon Generation Station on Lake Michigan dissolved oxygen and nitrogen levels were determined for water from the intake and discharge canals during January to March 1972. These data, in conjunction with published information from other sources, indicated that gas supersaturation levels as high as 130% have occurred (Otto 1972). However, he observed no symptoms in the fish collected from the discharge canal and concluded that the fish did not remain in the canal long enough to develop the disease. At sites where fish are more closely confined to the discharge area, there is a potential adverse impact due to supersaturation alone. For example, 13 species of warmwater fishes exhibited external symptoms of gas-bubble disease in the discharge canal and cove of a stream generating station. Distressed black crappie were observed to have gas emboli in the large vessels of the gill fragments (DeMont and Miller 1971). Later studies (Miller 1974) at the same site revealed nine species of fish with external symptoms of gas-bubble disease. Exophthalmus (pop-eye) of one or both eyes and gas bubbles in the fins were the most commonly observed symptoms. The disease incidence in all fishes captured peaked at 4.7% of the game species, white bass had the highest disease frequency (27.8%). Sampling of two other plants on nearby reservoirs demonstrated a very low incidence of gas-bubble disease (0.3% of total catch).

The principal point in the above discussion is that we should always attempt to define and describe all of the potential problems associated with a discharge. Not all stresses, such as infectious disease, gas-bubble disease, heat, and chemicals, will occur at all times, but without proper evaluation, these stresses could be overlooked.

ANALYTICAL AND CHEMICAL CONCERNS

The use of amperometric titration for the analysis of total residual chlorine is becoming the mainstay of regulatory agencies. In some situations there is a hesitancy about changing from various colorimetric procedures. The cost of instrumentation, the necessity of training laboratory or plant personnel, pure procuration, and other causes have delayed the use of this generally more sensitive and accurate method. There will be exceptions and it has been concluded by Nelson (1973) that the amperometric procedure for determining residual chlorine in aqueous solutions is not applicable to all cooling tower waters. Some cooling tower waters contain copper, turbidity, and water treatment chemicals. These constituents may produce interferences in the analytical procedure which result in erroneous residual chlorine readings. However, subsequent experimental results (Manabe 1974) have shown that the analytical difficulties in the application of the amperometric technique to cooling tower blowdown can be eliminated by the addition of a pyrophosphate reagent to the currently used amperometric procedure. Copper and iron concentrations in typical blowdown do not interfere in the presence of pyrophosphate.

The evaluation of chlorinated wastewater treatment plant effluent has led to another potential problem for the power generating industry. Jolley in two studies (1974, 1975) described techniques and experiments that separated over 40 ³⁶Cl-tagged chlorine-containing constituents in wastewater effluent chlorinated with ³⁶Cl-tagged chlorine gas or hypochlorite solution. He estimated that 907 metric tons of chlorine, in the form of stable chlorine-containing organic compounds, are released annually by wastewater treatment plants to receiving water ecosystems. Associated studies by Cichro et al. (1974) have evaluated the toxicity to carp eggs of two of these compounds formed, 4-chloro-2-methyl and 5-chloro-2-methyl.

Additional studies (Glase et al. 1973) have evaluated wastewater effluents before and after laboratory chlorination using sodium hypochlorite. They estimated that the observed analytical peaks represent concentrations of organic chlorine compounds in the range of 1-50 µg/l in the wastewater effluent.

Jolley et al. (1975) have recently applied these research tools to cooling water discharges. They collected unchlorinated effluent from the Kingston Steam Plant and chlorinated the sample in the laboratory using ³⁶Cl-tagged chlorine gas at a chlorine concentration of 2.5 mg/l for

75 minutes in order to determine whether chlorine-containing organic compounds are formed during chlorination. One of the results was that the total yield of chlorine as chloro-organics in the chlorination of the cooling water sample was 0.78% of the initial 3.1 mg/l chlorine dosage. A reasonable estimate based on actual chlorine usage at the Kingston Plant was about 0.3%. Based on an estimate of 80,000-100,000 tons of chlorine used annually in chlorinate cooling water, Bailey et al. (1975) concluded that several hundreds of tons of chlorinated organics are produced annually during chlorination of cooling systems.

RESEARCH NEEDS

Fortunately, several studies have begun, designed specifically to evaluate the real impact of chlorination of condenser effluent. The influence of water quality, especially as it influences the ratio of free to combined chlorine, needs further evaluation. The difficulties involved in maintaining and measuring different ratios to determine the magnitude of difference in toxicity have resulted in some confusion as to the need for distinct criteria for the two forms of chlorine. Charles Warren, Oregon State University, will be conducting side-by-side studies in which he will maintain a high percent occurrence of free chlorine in one system and added amounts of chloramines in another system. If a significant difference exists, intermediate ratios will be investigated. We will then also need a better knowledge of the occurrence of free chlorine in discharge plumes.

Temperature, as discussed earlier, has an inconsistent impact on toxicity. More detailed studies of this factor need to be conducted. If lower temperatures truly result in decreased toxicity then this fact could influence the chlorine criteria during winter months, during which time

we have the most significant problem due to attraction of fish to the warmer effluents.

Recent studies supported by the USEPA are showing that under acute continuous exposure to residual chlorine in wastewater effluent some species of minnows important as forage fish in lakes and rivers appear to be as sensitive as trout and salmon. Many more data need to be generated with a variety of important species to adequately protect the majority of these species. It could well be that the present distinction between coldwater and warmwater species is unjustified.

A distressed or disequibrated fish may not have time to recover in the areas of discharges due to predation or physical damage from velocity. Additional studies are needed in which distressed fish are observed for a few days after chlorination. In addition to using death as an observable biological endpoint, we also need an EC50 determination so we know what effective concentration results in a turnover (and possible eventual mortality) of 50% of the individuals.

It is quite important to study the exposure times and frequencies that actually occur. In freshwater, chlorination cycles frequently last for about 30 minutes and occur up to several times per day. Some of this chlorination has been shown to be excessive but it does provide a starting point for the design of experiments. Recent studies on the behavior and movement of fishes in and around heated plumes lead one to believe that these fish move about quite frequently and they may seldom be subjected to more than one chlorination. This impression must be evaluated to properly determine the number of chlorinations to which the test organisms are to be exposed.

Finally, we need more on-site studies of chlorination effects that, among other factors, will evaluate the ability of fish to avoid slugs of chlorine.

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